

Research Article – Environmental Biology

Phytotoxic Effect to Cockscomb (*Celosia cristata* L.) with Response of Chlorpyrifos Treatments**M. Santhoshkumar^a, L. Baskaran^{a*}, T. Mahakavi^a, R. Bakiyaraj^b and T. Ravimycin^a**^aDepartment of Botany, Annamalai University, Annamalai Nagar-608 002, Tamil Nadu, India^bDepartment of Botany, Government Arts College, Thanthonrimalai, Karur - 639 005, Tamil Nadu, India**Abstract**

The present work was conducted to evaluate the phytotoxic of chlorpyrifos on cockscomb (*Celosia cristata* L.) plant. The seeds were exposed to different dosages (control, 0.5, 1, 2 and 2.5%) of chlorpyrifos. On the 7th day seed germination percentage were determined, seedling morphological such as shoot length, root length, biomass and photosynthetic pigments like chlorophyll 'a' and chlorophyll 'b' were examined on 30, 60 and 90 days after treatments. The germination percentage negatively affected with increasing chlorpyrifos level. The morphological and photosynthetic pigments were increased at optimum level (0.5%). Moreover, above the optimum concentration all investigated parameters were decreased with increasing chlorpyrifos level. The obtained results suggest that usage of this insecticide optimum dosage helpful for good agricultural practices.

Key words: Pesticides, Chlorpyrifos, *Celosia cristata*, Phytotoxicity, Photosynthetic pigments

Introduction

Pesticide is specifically planned for the control of pests, weeds or diseases in modern agricultural practices. Application of pesticide is conceived the most effective and accepted means for the protection of plants from pest attack and has significantly brought to enhance agricultural productivity (Dubey *et al.*, 2015). Annually 4.6 million tons of chemical pesticide and about 500 types of pesticide were used across the world (Zhang *et al.*, 2011). Pesticides have become contaminated the soil, surface areas, ground water by the way of spills, accidents, misapplication, and/or runoff and soil erosion (Karthikayen *et al.*, 2004; Henderson *et al.*, 2006). Among the pesticides, the chlorpyrifos [O,O-diethyl O-(3,5,6-trichloro-2-pyridinyl)-phosphorothioate] is an organophosphorus insecticide extensively used to control pests on grain, fruit, cotton, nuts, vegetables crops, as well as lawns and ornamental plants. US-EPA (U.S. Environmental Protection Agency, 2002) has been confined for residential uses and even some agricultural purposes in United States and some European countries, but in India it was continuously used to control insects in agricultural crops (Eaton *et al.*, 2008).

In study of (Baig *et al.*, 2003) reported that application of glyphosate gradually declined the

germination of seed and shoot fresh weight in pea plant. In the experiment of Stevens *et al.*, 2008, studied that higher dose of imidacloprid significantly affected the germination of rice seedling. The study accounted that Pesticide can reduce the germination percentage and seedling height in barley (Srivastava and Singh, 2009). In earlier report (Kim and Ahn, 2009) suggest that excessive and frequent use of chlorpyrifos might have accumulated in agricultural crops. Previous result reported that chlorpyrifos toxicity in *Brassica chinensis* L. and concluded that chlorpyrifos significantly inhibit root growth (Zhi-Yong *et al.*, 2011). Earlier report (Basantani *et al.*, 2011) indicate that treatments of glyphosate importantly inhibit the germination percentage, fresh weight and root length of *Vigna radiata*. It has been earlier reported that dimethoate insecticide is toxic effect to the plant growth, photosynthetic pigments and photosynthetic activity in *Glycine max* L. (Panduranga *et al.*, 2005). Moreover, chlorpyrifos have negative impact on photosynthetic pigment, further increase of higher concentration (Kashyap and Kumar, 2013). Recently, Santhoshkumar *et al.*, 2015 examined the phytotoxicity of chlorpyrifos in green gram. In the present study, we have studied the effect of chlorpyrifos to establish the differential sensitivity of *Celosia cristata* L. against imposed pesticide. The study has following objectives as follows: (i) to detect the efficiency of the selected pesticide on morphological attributes such as germination percentage of seed, plant height and biomass. (ii) to study the relative effect of selected pesticide on photosynthetic pigments. To our

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knowledge the present investigation is the first report that phytotoxic effect of chlorpyrifos in Cockscomb (*Celosia cristata* L.) plant.

Materials and Methods

Chemicals and Plant material

The Chlorpyrifos 20% EC was purchased from agroagencies, chidamparam, India. Treatment concentrations were prepared from this solution as 0.5, 1, 2 and 2.5 %. The certified seeds of *Celosia cristata* were collected from the Department of Agriculture, Annamalai University, Annamalanagar, Tamil Nadu. Seeds were surface sterilized with 0.5% sodium hypochloride for 10 min, followed by extensive washing with sterile distilled water. The seeds were placed on two layered moist filter paper in petriplates and placed in an incubator to germinate at 25°C for 7 days. The emergence of radicle was considered for seed germination. The total germination percentage was calculated by using the formula.

Germination percentage = Total number of seeds germinated/ Total number of seeds sown \times 100

Morphological parameters

The morphological parameters including shoot length, fresh weight and dry weight were analysed at the periods of 30, 60 and 90 days after sowing (DAS). For the measurement of shoot and root length, twenty plants were randomly selected from each treatment to record the seedling growth. The growth was measured using a centimeter scale and the values were recorded. For the measure of fresh weight and dry weight the plants sample were kept in an hot air oven at 80°C for 24 h. Then, the samples were kept in desiccators and their dry weight was recorded by using an electrical single pan balance. The average was expressed in g⁻¹ plant.

Photosynthetic pigments estimation

The photosynthetic pigments chlorophyll 'a', chlorophyll 'b' with 30, 60 and 90 days old *Celosia cristata* was estimated followed by Arnon, (1949) method. Five hundred mg of fresh leaf material was ground in a mortar and pestle with 10 mL of 80 per cent acetone. The homogenate was centrifuged at 800 rpm for 15 min. The supernatant was saved. The residue was re-extracted with 10 mL of 80 per cent acetone. The supernatant was saved and the absorbance values were read at 645 and 663 nm using visible UV-Spectrophotometer (Hitachi). The chlorophyll 'a', chlorophyll 'b' and total chlorophyll contents were estimated and expressed in mg g⁻¹ fresh weight basis.

Chlorophyll 'a' = (0.0127) \times (O.D 663) – (0.00269) \times (O.D 645)

Chlorophyll 'b' = (0.0229) \times (O.D 645) – (0.00488) \times (O.D 663)

Statistical analysis

All treatments were replicated three times. All presented data are expressed as mean \pm standard deviation (SD) of the mean. The statistical graphs were carried out by using Microsoft offices excel.

Results

Effects on seed germination percentage

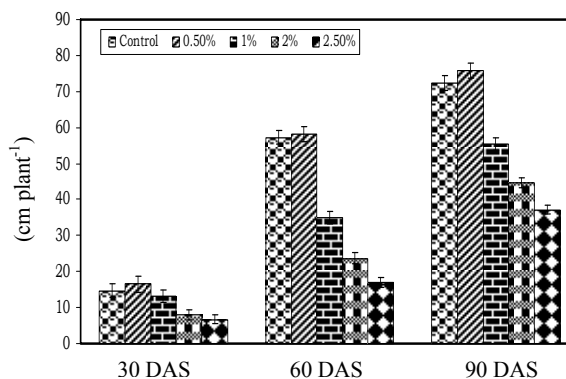
Germination rate was decreased by 80, 66, 53 and 46 % at 0.5, 1, 2 and 2.5%, respectively. The most effect (46%) of chlorpyrifos on seed germination was found at the highest concentration 2.5%.

Effect on morphological parameters

The effect of different concentrations of chlorpyrifos on shoot length shown in Fig-1. The shoot length was increased at 0.5% treatment concentration with comparison of 16.5, 58.2 and 75.8 cm respectively, which was compared with control 14.7, 57.2 and 72.3 cm. Thereafter, the shoot length was decreased with increasing the concentrations. The most destructive effect on shoot length (6.7, 17 and 37.2 cm) was observed in 2.5% concentration for all the treatments days.

The effect of root length of chlorpyrifos are depicted in Fig-2. The highest reduction of root length (4.3, 4.7 and 7.4 cm) was observed at the higher concentration of treatments. The fresh weight of plant is shown in F-3. Increasing concentration of chlorpyrifos affect the fresh weight, the most destructive effect was observed in 2.5% concentration in all the treatments days 2.08, 12.95 and 12.94 g/plant respectively. Dry weight was gradually declined with increase of chlorpyrifos dosage level 0.42, 1.95 and 0.98 g/plant, respectively (Fig-4).

Fig-1. An influence of different concentrations of chlorpyrifos on shoot length



Influence of chlorpyrifos on chlorophyll content

The photosynthetic pigment such as chlorophyll 'a' and chlorophyll 'b' after the 30, 60

and 90 days, respective treatments are given in Fig-5 and Fig-6. Plant treated with 0.5% of chlorpyrifos exhibited an increase of in the value of chl *a* and chl *b* (3.16, 5.92, 4.6 and 1.93, 2.86, 2.18 mg g⁻¹ fr. wt) respectively. In contrast, decrease in chl *a* and chl *b* content was observed in higher concentration (1.45, 3.64, 2.98 and 0.91, 1.95, 0.98 mg g⁻¹ fr. wt) respectively.

Fig-2. An influence of different concentrations of chlorpyrifos on root length

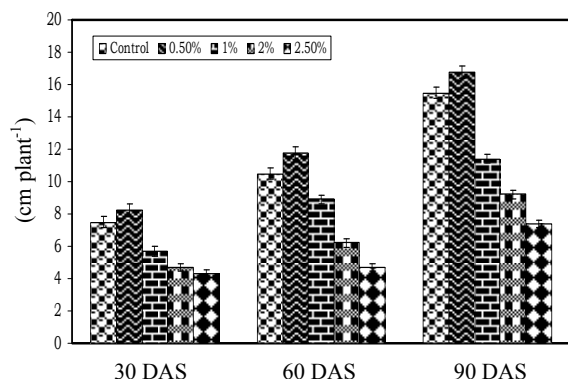


Fig-3. An influence of different concentrations of chlorpyrifos on fresh weight

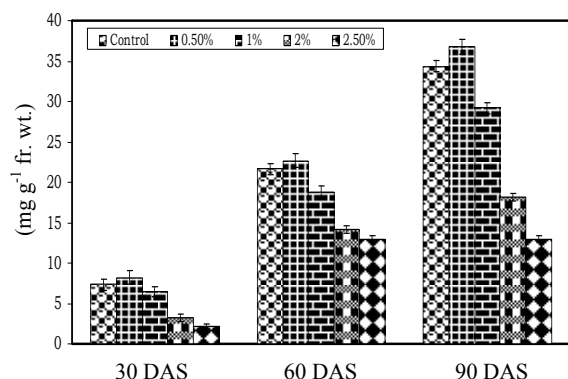
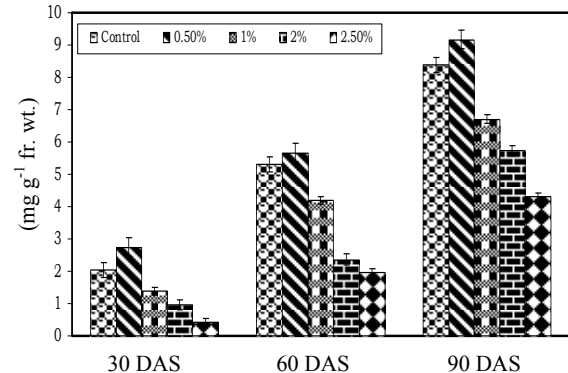


Fig-4. An influence of different concentrations of chlorpyrifos on dry weight



Discussion

Plant growth parameters

The plant growth parameters such as shoot length, root length and biomass (fresh weight and

dry weight) was increased at 0.5% of chlorpyrifos when compare to control plants. In contrast, higher concentrations of chlorpyrifos, a declined in above parameters were recorded in all the observed days. Seedling growth prohibition might be due to the inhibition of hydrolytic enzyme synthesis or blocking of enzyme pathway in seed during germination (Gange *et al.*, 1992).

Fig-5. An influence of different concentrations of chlorpyrifos on chl *a*

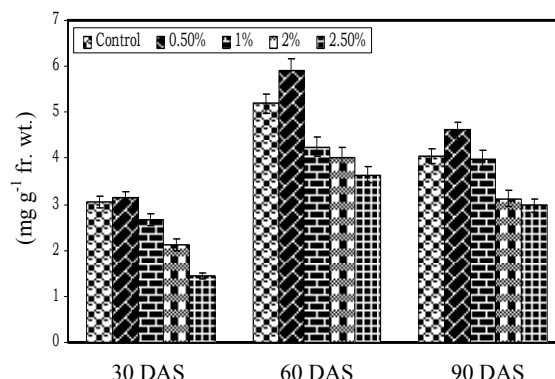
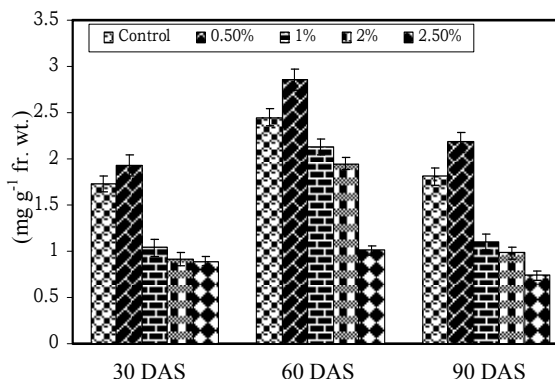


Fig-6. An influence of different concentrations of chlorpyrifos on chl *b*



Under abiotic stress including pesticide, the plant activate and formed some phenolic compounds like isoflavones (genistein, diadzein) phenolic acid (elagic, tannic and vanilic acid) and hydroxycinnamic acid derivatives (ferulic acid, p-hydroxy benzoic acid and p-caumeric acid), could reduce the germination and plant growth (Macias *et al.*, 1992; Mersie and Singh, 1993). Moreover, seed germination is an crucial phase in plant growth, and especially sensitive to stress tolerance against contaminants (Banks and Schultz, 2005). Seed germination percentage may reflect the plants seeds on their living environment (Li *et al.*, 2007). However, overall inhibition in seedling and morphological parameters was observed in *V.mungo* with the application endosulfan (Mishra and Mohanty, 2008). The presence of pesticide residue in soil could inhibit the uptake of micro nutrients and create nutrient deficiency, this may reflect in abnormality in plant growth parameter (Wahengbam *et al.*, 2013).

Recently Bassey *et al.*, 2015 studied the seed germination of soyabean (*Glycin max*) in the treatments of chlorpyrifos and ridomil. They found that seed germination was declined over treatment of pesticides.

Plant morphology is connected with cell division and cell elongation or the conversion of indole-3 acetic acid into different photo oxidative products (Tevivi and Teramura, 1989; Ros and Tevivi, 1995). The shoot and root dry mass was decreased in faba bean when grown in brominal and gramoxone treated soil, even at field condition (Abd-alla and Omar, 1993). On other hand, root and shoot dry weight was declined over the treatment of glyphosate on soybean concluded by King *et al.*, 2001. Insecticide inhibit the plant height with increase of concentration, it might be due to retarded cell growth and division in root Warabi *et al.*, 2001. Liu *et al.*, 2009 observed that the root elongation was decreased with the increasing concentration of pesticide of cypermerthrin in Chinese cabbage (pakchoi) seed. Decrease of root and shoot length with the treatment of dimethoate may be due to arrest of physiological and biochemical process (Mishra *et al.*, 2009). Similar result was observed (Prasertsup and Ariyakanon, 2011) that plant biomass was increased in lowest concentration of chlorpyrifos, but higher concentration decreased in plant biomass in both *Pistia stratiotes* and *L. minor*. As the increase of deltamethrin inhibit the radicle length of root is likely relation to the abnormality of chromosome (Duran *et al.*, 2015).

Photosynthetic pigment content

Photosynthetic pigments content such as chlorophyll 'a' and chlorophyll 'b' exhibited an increase for 0.5% of chlorpyrifos after all the treatments days over the control plant, but with increase in the concentrations of chlorpyrifos, all the used concentrations of chlorpyrifos inhibit the pigment content. Vermass, 1993 ;Feurtet-Mazel *et al.*, 1996 concluded that phytotoxic effect is depend on the inhibition of photo dependent electron transport in thylakoids, at level of photosystem II (PS II). Sresen *et al.*, 2000 stated that the reduction of chlorophyll content is might be decrease of leaf area with increasing pesticide. Tort and Turkyilmaz, 2003 noted that the application fungicide reduced chl *a, b* and total chlorophyll in pepper leaves with higher dose of fungicide. Mishra *et al.*, 2008 reported that the insecticide could affect photosynthetic system by the inhibition of photo system II and electron chain transport activities. Abhilash and sikh, 2010 stated that the organic pollutant can change the lipid composition of the chloroplast by affecting the photosynthetic pigments and key enzymes. Raja *et al.*, 2012 described that when increase of pesticide concentration the chlorophyll-a, chlorophyll-b and total chlorophyll content was progressively diminished in *Azolla*

microphylla. Similar result observed by (Padhy and panigrahi, 2015) described that lower concentration of pesticide increase chlorophyll content whereas, higher concentration of pesticide reduced chlorophyll in *Anabaena cylindrica* Lemm.

Conclusion

The present studied to assess the effect of chlorpyrifos treatment on seed germination and seedling growth parameters include shoot length, root length, biomass and photosynthetic pigment. The result of this investigation shows that optimum concentration increase all the investigated parameters. While seed germination, seedling parameters as well as photosynthetic (chl *a*, chl *b*) were adversely affect with increasing concentrations. The study suggest that chlorpyrifos is broadly used insecticide, usage of this insecticide optimum dosage helpful for good agricultural practices. This study will help to determine phytoremediation potential of cockscomb plant with response chlorpyrifos contaminated soil.

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